DC potentials for these studies were as follows: initial gradient potential (electrode #1), 225 V; final gradient potential (electrode #25), 80 V; electrode #26, 70 V; electrode #27, 50 V; electrode #28, 25 V; final oriface electrode, 10 V.

The current transmitted to the octapole ion guide was measured by tightly covering the entrance to the octapole with aluminum foil and then measuring the current with a Keithley (Model 617) picoammeter. Ion current entering the mass spectrometer was measured using the picoammeter via a nickel coated brass plate (38 mm o.d.) located approximately 5 mm beyond the exit of the heated capillary inlet.

Electrospray emitter "tips" were made by pulling 0.185 mm o.d., 0.050 mm i.d. fused silica capillary tubing (Polymicro Technologies, Phoenix, Ariz., USA). The electrospray voltage was 2.0 kV and the capillary inlet was biased at 500 volts (ion funnel interface only) using DC power supplies (Models 305 and 303, respectively, Bertan, Hicksville, N.Y., USA). Mass spectra and ion current measurements were obtained at an ESI flow rate of either 200 or 400 nL/min using a Harvard syringe pump (South Natick, Mass., USA). The heated capillary inlet was maintained at a temperature between 170–215° C. The ion funnel was operated at a frequency of 700 kHz or as otherwise indicated.

For comparison, mass spectra were acquired using the standard TSQ 7000 ESI ion source equipped with a 114 mm long and 0.41 mm i.d. heated capillary inlet using similar operating and tuning conditions to that used with the ion funnel. The mass spectra obtained with the standard ESI ion source were measured with three different Finnigan capillary inlets (identical dimensions) for the data presented (e.g. reconstructed ion currents). In either the case of the ion funnel or standard ESI ion source, the mass spectrometer was tuned to maximize ion transmission and obtain identical resolution for selected peaks from a 2.9 M solution of horse heart myoglobin or a mixture containing 2.9 M of horse heart myoglobin and 20.0 M synthetic Phe-Met-Arg-Phe amide, depending on the required mass range. Conditions such as electrospray voltage (2.0 kV), capillary inlet temperature (200 ° C.), electron multiplier voltage (1200 or 1400 V), sample flow rate (200 or 400 nL/min), acquisition scan rate (typically m/z 200–2500 in 3 seconds), and total acquisition time (1 or 2 min. averages) were held constant when directly comparing spectra from the two designs. The ion source block was pumped by an Edwards (Wilmington, Mass., USA) mechanical pump (549 L/min). The pressure measured in ion source block (i.e. between the capillary inlet and the skimmer) was 870–915 mTorr and in the region of the mass analyzer was ~2–4×10^{-6} Torr. All of the data presented was reproduced at least twice.

Myoglobin (horse heart), cytochrome c (horse heart), ubiquitin (bovine red blood cells), gramicidin S (bacillus brevis, hydrochloride salt), Phe-Met-Arg-Phe amide (synthetic), polyethylene glycol (avg. mol. weight, 8000 amu), methanol, and glacial acetic acid were purchased from Sigma (St. Louis, Mo., USA). Standard solutions were prepared in methanol/deionized water/acetic acid (50:50:1%) except for polyethylene glycol which was prepared in methanol/deionized water (50:50). Solutions were kept refrigerated and were prepared from the corresponding standard material biweekly or as needed.

Results and Discussion

The purpose of this embodiment of the ion funnel interface is to realize improved sensitivity by more efficient transmission of the electrospray ion current to the mass analyzer. The ion funnels ability to do this rests upon three aspects of operation: (a) efficient capture of the electrospray ion plume emanating from the heated capillary, (b) effective collisional focusing of the ions in the ion funnel through the use of RF fields and (c) the imposed drift of the ions towards the bottom, or exit, of the funnel due to the DC potential gradient. The observed results, in terms of ion current measurements and mass spectra, supported these basic premises.

Ion Current Measurements. Initial experiments involved measuring ESI current collected on a plate at ground immediately following the final electrode of the ion funnel. FIG. **7**, data set A, shows a plot of detected current measured for the 100–400 V_{pp} RF amplitude range from ESI of a 58 μM bovine ubiquitin solution. Beginning at 15 pA, corresponding to DC-only mode of operation, the detected ion current increases as the RF amplitude was increased to a maximum exceeding 1800 pA. This two order of magnitude increase in detected current demonstrates that the presence of RF fields with this device clearly results in improved ion focusing. The effects of RF fields at the bottom of the funnel were explored in particular because it is a region where space-charge and other effects are likely to be most problematic. Using the adjustable RF/DC coupler, the RF amplitude on electrodes #26–28 were reduced relative to the nominal RF amplitude on electrodes #1–25. It is noteable that the change in operation frequency from 700 to 825 kHz reflects the change in resonating frequency of the series circuit (i.e. the adjustable RF/DC circuit, high-Q-head, and ion funnel). An ESI of the same ubiquitin solution and operating at 80% and 0% of the nominal RF amplitude applied to electrodes #1–25 yielded a maximum ion current of 1.1 and 0.5 nA, respectively FIG. **7**, data set B and C respectively. The overall shape of these two curves are similar but the overall amount of detected ion current was reduced to less than half by operating ion funnel electrodes #26–28 in the DC-only mode. Interestingly, the shape of the curve at 700 kHz is markedly different and shows a much sharper transmission maximum than the curves taken at 825 kHz. Thus, the data shows that the RF fields clearly mediate the ion current focused through the interface and that the presence of RF fields in the bottom of the funnel effect ion transmission through the ion funnel device.

To accomplish effective capture of the expanding ion plume, the exit of the heated capillary was positioned so as to be both flush with the opening of the first electrode and aligned with the central axis of the funnel. This choice was based in part on results that indicated maximum ion currents (58 μM ubiquitin solution) detected when the heated capillary was flush with the opening of the first electrode. Secondly, the heated capillary inlet was maintained at a higher relative potential to that of electrode #1, thus ensuring the ions movement into the entrance of the ion funnel. For example, for positive ions, with the initial potential of the DC gradient on electrode #1 set at 300 V, ion transmission (same ubiquitin solution) was consistent for a heated capillary inlet potential in the 300–500 V range. However, if the capillary potential was lowered to 200 V then the observed transmission in ion current decreased to approximately 70% of the values observed for a capillary voltage in the 300–500 V range. The latter observation corresponds to a fraction of the ions electrostatically rejected from entering the funnel.

Ion currents were also measured as a function of concentration for ubiquitin solutions ranging from 0.58 to 58 M (FIG. **7**, data sets A, D, and E respectively). The detected current increased (although not linearly) with the concentration of the analyte. This indicates that the majority of the